(19) Canadian Intellectual Property Office
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Un organisme d'Industrie Canada

(11) CA 2 310 700 (13) A1

(40) 03.06.2001
(43) 03.06.2001

(12)

(21) 2 310 700

(22) 06.06.2000

(51) Int. Cl.7: B81B 1/00, B81C 1/00, B01D 57/02

(30) 11-345050 JP 03.12.1999

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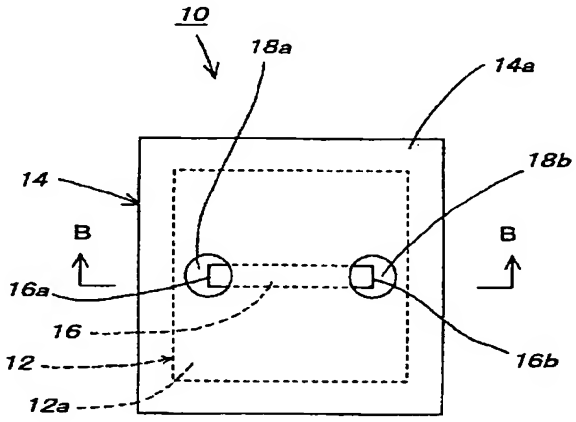
(54) PUCE ELECTRONIQUE POUR ELECTROPHORESE CAPILLAIRE EN GEL, ET METHODE DE FABRICATION CONNEXE

(54) MICROCHIP FOR CAPILLARY GEL ELECTROPHORESIS AND THE METHOD FOR FABRICATING THE SAME

(57)

In order to provide a microchip for capillary gel electrophoresis which can be fabricated at low cost and is suitable for disposable use wherein it is discarded after having been used only one time for electrophoretic separations, the microchip is composed of a planar substrate prepared from a polymer material and a planar surface plate to be disposed on the top surface of the substrate, a capillary channel constituting a flow path having a predetermined contour being defined on the top surface of the substrate, and the capillary channel being sealed with the surface plate.



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CIPO
CANADIAN INTELLECTUAL PROPERTY OFFICE

(12)(19)(CA) Demande-Application

(21)(A1) 2,310,700

(22) 2000/06/06
(43) 2001/06/03

(72) HONG, JONG WOOK, JP
(72) FUJII, TERUO, JP
(72) SEKI, MINORU, JP
(72) ENDO, ISAO, JP
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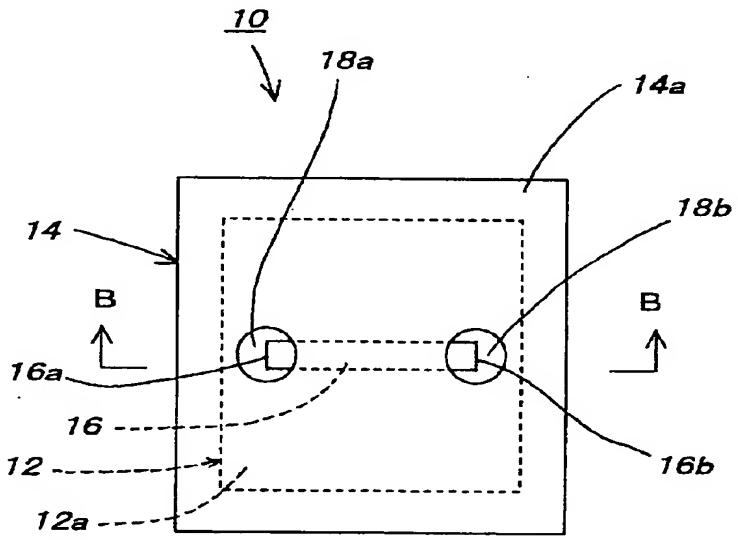
(51) Int.Cl.[7] B81B 1/00, B01D 57/02, B81C 1/00
(30) 1999/12/03 (11-345050) JP

(54) PUCE ELECTRONIQUE POUR ELECTROPHORESE CAPILLAIRE EN GEL, ET METHODE DE FABRICATION CONNEXE

(54) MICROCHIP FOR CAPILLARY GEL ELECTROPHORESIS AND THE METHOD FOR FABRICATING THE SAME



(57) In order to provide a microchip for capillary gel electrophoresis which can be fabricated at low cost and is suitable for disposable use wherein it is discarded after having been used only one time for electrophoretic separations, the microchip is composed of a planar substrate prepared from a polymer material and a planar surface plate to be disposed on the top surface of the substrate, a capillary channel constituting a flow path having a predetermined contour being defined on the top surface of the substrate, and the capillary channel being sealed with the surface plate.

Abstract of The Disclosure

In order to provide a microchip for capillary gel electrophoresis which can be fabricated at low cost and is suitable for disposable use wherein it is discarded after having been used only one time for electrophoretic separations, the microchip is composed of a planar substrate prepared from a polymer material and a planar surface plate to be disposed on the top surface of the substrate, a capillary channel constituting a flow path having a predetermined contour being defined on the top surface of the substrate, and the capillary channel being sealed with the surface plate.

SPECIFICATION

Title of the Invention

MICROCHIP FOR CAPILLARY GEL ELECTROPHORESIS AND THE METHOD FOR FABRICATING THE SAME

Background of the Invention

Field of The Invention

The present invention relates to a microchip for capillary gel electrophoresis and the method for fabricating the same, and more particularly to a microchip for capillary gel electrophoresis used suitably for separating nucleic acids including a variety of sizes of DNA fractions; organic molecules such as amino acids, peptides, and proteins; metal ions and the like in a small experimental scale; as well as to the method for fabricating the same.

Description of The Related Art

Heretofore, it has been known that a capillary channel is defined on a chip composed of planar glass by means of microfabrication in accordance with photolithographic technique to constitute a chip for capillary electrophoresis, and that when the chip for capillary electrophoresis thus fabricated is used, it becomes possible to perform high-quality and high-speed electrophoretic separations.

In these circumstances, many researches have been made with respect to the above described chip for capillary electrophoresis wherein a chip prepared from a silicon material

such as glass, and Si/SiO_2 is employed, and a capillary channel is defined thereon in accordance with microfabrication.

In this respect, however, etching and bonding processes having complicated operational steps must be carried out for forming a capillary channel used in electrophoretic separations, and for sealing positively the resulting capillary channel in the case where glass or silicon is used as a material of a chip for capillary electrophoresis.

Accordingly, a chip for capillary electrophoresis which is prepared from a glass material or a silicon material becomes inevitably expensive as a result of increasing its manufacturing cost. Thus, discarding of such microchip as described above for electrophoretic separations after only one time application thereof did not pay from the viewpoint of cost.

For this reason, in a chip for capillary electrophoresis wherein glass or silicon is used as its material, such a gel which is difficult to remove from a capillary channel after having been filled therewith is not employed as a separation material (molecular sieves) to be filled into the capillary channel for the sake of making the same possible to use in repeating electrophoretic separations, but it is necessary for using a buffer solution which can be allowed to freely flow away from a capillary channel after having been filled therewith, or an easily replaceable high molecular (polymer) solution such as linear polyacrylamide, and hydroxypropyl cellulose, so that an electrophoretic separation has been carried out by the use of these molecular sieves.

However, in the case where electrophoretic separations are carried out by using a buffer solution or a polymer solution as molecular sieves in a chip for capillary electrophoresis prepared from a glass or silicon material, it is required to use high voltage as well as to control delicately an electric field in order to prevent occurrence of diffusion or convection which arises in application of voltage. Accordingly, there is such a problem that electrical equipment and detection devices become complicated, resulting in high cost, and further there is also such a problem that since a long capillary channel is necessary for increasing a degree of separation, a chip must be inevitably large-sized.

Objects and Summary of The Invention

The present invention has been made in view of a variety of problems involved in the prior art as described above, and an object of the invention is to provide a disposable microchip for capillary gel electrophoresis which can be fabricated in low cost and suitable for discarding thereof after having been only once used for electrophoresis as well as to provide a method for fabricating the same.

Another object of the present invention is to provide a microchip for capillary gel electrophoresis by which it becomes possible to use a gel as molecular sieves, whereby electrophoretic separations can be effected with low voltage, but not high voltage, and further occurrences of diffusion and convection at the time of applying an voltage are suppressed, whereby a simplification of electrical equipment and detection

devices is intended, so that it becomes possible to remarkably reduce its cost as well as to provide a method for fabricating the same.

A still further object of the present invention is to provide a microchip for capillary gel electrophoresis by which it becomes possible to use a gel as molecular sieves, whereby it is intended to improve a degree of separation in electrophoretic separations, reduction of a separation distance, and reduction of a separation time, so that it becomes possible to improve separating performance in electrophoretic separations and to effect electrophoretic separations in high separation resolution as well as to provide a method for fabricating the same.

In order to achieve the above described objects, according to the present invention, minute processing is applied to a microchip prepared from a high-molecular (polymer) material such as PDMS (polydimethyl siloxane), and a capillary channel is defined on the microchip.

In this case, although silicon such as glass and Si/SiO_2 has been used heretofore as a material of microchip for defining a capillary channel in accordance with minute processing, it is preferred to use a polymer material in view of low cost as compared with that of glass or Si/SiO_2, besides the latter is less fragile than the former.

Particularly, PDMS being a kind of silicone elastomer included in polymer materials is a material used suitably for molding in microscale, by the use of which a capillary channel

being a microstructure for electrophoretic separations can be processed and molded in low cost.

In other words, when a microchip prepared from PDMS is employed in case of forming capillary channels, such capillary channels can be easily formed in accordance with simple and inexpensive molding and sealing manners without accompanying such etching and bonding processes which require complicated operations.

As a result, a microchip for capillary gel electrophoresis according to the present invention can be inexpensively provided, so that it becomes possible to apply a so-called disposable use in which a microchip is discarded after it had been once used in an electrophoretic separation by the use of the microchip for capillary gel electrophoresis of the present invention.

In addition, when a microchip is disposable as described above, it is possible to use no buffer solution or no polymer solution which can be removed from a capillary channel after having been filled therewith as molecular sieves being a separation material with which the capillary channel is to be filled.

For this reason, in the present invention, a capillary channel defined on a microchip for capillary gel electrophoresis is filled partially with a gel (for example, agarose gel), whereby separation resolution can be improved as compared with electrophoretic separations carried out on a microchip wherein a buffer solution or a polymer solution, which has been heretofore used as molecular sieves, is employed,

besides a longer length of the capillary channel required for separations can be reduced.

Thus, according to a microchip for capillary gel electrophoresis of the present invention, since a gel may be used as molecular sieves to be filled in a capillary channel, it becomes possible to effect electrophoretic separations with high resolution without requiring extension of a capillary channel and control of a delicate electric field for the sake of suppressing problems of occurring diffusion and convection.

As a result, it becomes possible to separate DNA molecules having a variety of sizes with simple equipment by the use of a microchip for capillary gel electrophoresis prepared from inexpensive PDMS for electrophoretic separations in accordance with the present invention.

As mentioned hereinafter, it has been succeeded in experiments by the present applicant that DNA molecules are electrically separated in reality on a microchip for capillary gel electrophoresis prepared from PDMS according to the present invention, whereby a band corresponding to molecular weight is formed. More specifically, in the experiments by the present applicant which will be described hereunder, agarose is first employed as a gel for molecular sieves in order to exhibit suitability of a microchip for capillary gel electrophoresis prepared from PDMS according to the present invention, and DNA ladders labelled with FITC are separated.

More specifically, a microchip for capillary gel electrophoresis of the present invention is composed of a planar substrate prepared from a polymer material and a planar surface plate to be disposed on the top surface of the substrate, a capillary channel constituting a flow path having a predetermined contour being defined on the top surface of the substrate, and the capillary channel being sealed with the surface plate.

In this case, the surface plate may be prepared from, for example, PMMA (polymethyl methacrylate), PDMS (polydimethyl siloxane), glass or the like.

On one hand, the substrate may be prepared from, for example, PDMS (polydimethyl siloxane).

Furthermore, a surface of the capillary channel defined on the top surface of the substrate may be made, for example, to be hydrophilic.

Moreover, in case of making the surface of the capillary channel defined on the top surface of the substrate hydrophilic, it may be made the surface of the capillary channel defined on the top surface of the substrate hydrophilic by means of, for example, oxidation with oxygen plasma.

On the other hand, a method for fabricating a microchip for capillary gel electrophoresis of the present invention comprises a first treatment for printing a layout pattern of a capillary channel in a microchip for capillary gel electrophoresis on a transparent film to prepare a photolithographic mask; a second treatment for forming a

negative photoresist on a silicon wafer; a third treatment for transferring the layout pattern printed on the mask prepared by the aforesaid first treatment to the negative photoresist prepared by the aforesaid second treatment, and developing the same to prepare a master; a fourth treatment for treating the master prepared by the aforesaid third treatment with fluorocarbon; a fifth treatment for pouring a mixed solution consisting of a PDMS prepolymer and a curing agent over the master treated with fluorocarbon in the aforesaid fourth treatment, and curing the same at a predetermined temperature for a predetermined period of time; a sixth treatment for peeling a PDMS replica away from the master after completing the curing for a predetermined period of time in the aforesaid fifth treatment, so that the PDMS replica is obtained as a substrate on which a capillary channel has been defined; a seventh treatment for covering the substrate obtained in the aforesaid sixth treatment by a surface plate, and attaching the latter to the former to seal the substrate with the surface plate; and an eighth treatment for making a surface of the capillary channel defined on the PDMS substrate to which the surface plate has been attached hydrophilic.

The treatment for making the surface of the capillary channel defined on the PDMS substrate in the aforesaid eighth treatment hydrophilic may be, for example, the one wherein the PDMS substrate to which the surface plate has been attached in the aforesaid seventh treatment is oxidized with oxygen plasma, so that the capillary channel defined on the substrate is oxidized with oxygen plasma, whereby the surface of the

capillary channel is made to be hydrophilic.

With respect to the above described hydrophilic treatment, other manners for achieving hydrophilic treatment than the oxidation with oxygen plasma as described above may be suitably utilized.

Brief Description of The Drawings

The present invention will become more fully understood from the detailed description given hereinafter and the accompanying drawings which are given by way of illustration only, and thus are not limitative of the present invention, and wherein:

FIGS. 1(a) and (b) show an example of a preferred embodiment of a microchip for capillary gel electrophoresis according to the present invention wherein FIG. 1(a) is a schematic view taken in the direction of the arrow along the line A of FIG. 1(b), and FIG. 1(b) is a sectional view taken along the line B-B of FIG. 1(a);

FIGS. 2 (a), (b), (c), (d) and (e) are schematic explanatory views each illustrating a process for fabricating a microchip 10 for capillary gel electrophoresis;

FIG. 3 is a constitutional explanatory view showing a system for detecting fluorescence in DNA labelled with FITC;

FIG. 4 is a scanning electron micrograph of a capillary channel defined on a substrate of the microchip for capillary gel electrophoresis shown in FIG. 1;

FIGS. 5(A), (B) and (C) are micrographs each showing a

situation of introduction and separation of DNA molecules;

FIG. 6 is a graphical representation indicating changes in electrophoresis of a microchip for capillary gel with time wherein a capillary channel 16 is filled with agarose gel containing DNA size standards of 100 bp to 1000 bp; and

FIG. 7 is a scanning electron micrograph of a capillary channel having another shape defined on a substrate of a microchip for capillary gel electrophoresis.

Detailed Description of The Preferred Embodiments

An example of a preferred embodiment of a microchip for capillary gel electrophoresis as well as a method for fabricating the same according to the present invention will be described in detail hereinafter in conjunction with the accompanying drawings.

FIGS. 1(a) and (b) show an example of a preferred embodiment of a microchip for capillary gel electrophoresis according to the present invention wherein FIG. 1(a) is a schematic view taken in the direction of the arrow along the line A of FIG. 1(b), and FIG. 1(b) is a sectional view taken along the line B-B of FIG. 1(a).

In these figures, a microchip 10 for capillary gel electrophoresis is composed of a planar substrate 12 prepared from PDMS, and a planar surface plate 14 prepared from PMMA (polymethyl methacrylate) which is disposed on the top surface 12a of the substrate 12.

Further, a capillary channel 16 is formed on the top surface 12a of the substrate 12 so as to define a so-called I-shaped flow pass.

Namely, the capillary channel 16 defined on the top surface 12a of the substrate 12 is sealed with the surface plate 14.

Two ports 18a and 18b being openings each of which is formed so as to pass through a bottom surface 14b from the top surface 14a of the surface plate 14 are bored on the surface plate 14 for introducing a sample and mounting electrodes, respectively.

In this case, two ports 18a and 18b as well as the capillary channel 16 are defined and arranged to have each size in such that opposite ends 16a and 16b of the capillary channel 16 are located over a part of each port 18a or 18b so that the port 18a and the port 18b are communicated with the end 16a and the end 16b, respectively.

Moreover, a length of the capillary channel 16 is determined, for example, to be 14 mm, a width of the capillary channel 16 is determined, for example, to be 400 μ m, and a depth of the capillary channel 16 is determined, for example, to be 40 μ m, respectively.

It is to be noted that a length of the capillary channel 16 is not specifically limited, but it may be arbitrarily determined according to need, a width of the capillary channel 16 is also not particularly restricted, but it may be arbitrarily determined as occasion demands, for example, it may be determined to be any optional value ranging from 10 μ m to 800

μ m, and a depth of the capillary channel 16 is not specifically limited, but it may arbitrarily be determined as circumstances demand, for example, it may be determined to be any arbitrary value ranging from 5 μ m to 150 μ m.

A fabricating process which will be described by referring to FIGS. 2(a), (b), (c), (d) and (e) is applied for fabricating the above described microchip 10 for capillary gel electrophoresis. In this respect, first, a layout pattern of the capillary channel 16 in the microchip 10 for capillary gel electrophoresis has been printed on a transparent film at high resolution, e.g., 4064 dpi prior to the fabricating process in order that the printed layout pattern is utilized as a mask for photolithography.

In the following, a process for fabricating the microchip 10 for capillary gel electrophoresis containing the above described substrate 12 prepared from PDMS will be described in detail.

An outline of a fabrication process of the microchip 10 for capillary gel electrophoresis is illustrated in FIGS. 2(a), (b), (c), (d) and (e).

A 20 mm x 20 mm silicon (Si) wafer is dried in an oven (FIG. 2(a)), and the negative photoresist SU-8 is spin-coated at 2,500 rpm for 20 seconds, thereafter the resulting photoresist is baked in the oven at 90°C for 30 minutes (FIG. 2(b)).

In the present embodiment, such a process wherein the negative photoresist SU-8 is spin-coated at 2,500 rpm for 20 seconds, and baked in the oven at 90°C for 30 minutes was repeated

twice or so in order to prepare a capillary channel structure having 40 μ m depth.

Then, the layout pattern in the microchip 10 for capillary gel electrophoresis which has been printed on a mask was transferred to a silicon wafer coated with SU-8 in accordance with a photolithographical manner by the use of a mask aligner (for example, "PEM-800; Union Optical Co., Tokyo, Japan" may be employed as a mask aligner), and the resulting material was developed in 1-methoxy-2-propyl acetate for 20 minutes (FIG. 2(c)).

The master thus fabricated was washed in isopropyl alcohol, and consecutively distilled water.

The master had been treated with fluorocarbon by the use of RIE (Reactive Ion Etching) system before pouring a PDMS prepolymer.

The fluorocarbon treatment is useful for removing the PDMS replica after molding.

Then, the PDMS prepolymer and a curing agent (for example, "Sylgard 184": Dow Corning Co., MI may be used as a curing agent) were admixed at a ratio of 10 : 1, the admixture was stirred thoroughly, and then, degassed in vacuum for only 15 minutes to prepare a prepolymer mixed solution. The prepolymer mixed solution thus prepared was poured over the master and cured at 65°C for 1 hour, thereafter 135°C for 15 minutes (FIG. 2(d)).

After the above described curing, when the PDMS replica was peeled off from the master, the PDMS substrate 12 was obtained. Then, the PDMS substrate 12 was attached to the PMMA

surface plate 14 on which had been bored the ports 18a and 18b so as to cover the PMMA surface plate 14, whereby the capillary channel 16 (FIG. 2(e)) is sealed therewith.

In the present preferred embodiment, an expression "to seal the capillary channel 16" does not mean to seal completely the capillary channel 16, but to arrange the opposite ends 16a and 16b of the capillary channel 16 as well as two ports 18a and 18b in such that the former members communicate with the latter members, respectively.

Furthermore, the PDMS substrate 12 attached to the PMMA surface plate 14 was oxidized with oxygen plasma by employing RIE system, whereby the capillary channel 16 was oxidized with oxygen plasma to achieve surface hydrophilicity of the capillary channel 16.

It is to be noted that a manner for achieving surface hydrophilicity of the capillary channel 16 is not limited to the above described oxidizing manner with oxygen plasma, but the other manners may suitably be utilized.

In the following, a preparation of a gel used for electrophoretic separations in the microchip 10 for capillary gel electrophoresis will be described.

First, agarose powder (for example, "SeaKem GTG agarose; FMC BioProducts, ME" may be employed as such agarose powder) was dissolved into 1 time larger volume of TBE (tris borate EDTA) buffer solution while heating them in an oven to prepare an agarose solution. The resulting agarose solution was kept in the oven at 65°C, then, it was introduced into the capillary

channel 16 from the ports 18a and 18b in the microchip 10 for capillary gel electrophoresis by utilizing capillary action, and the resulting member was allowed to stand at room temperature for 5 minutes to cure the agarose solution.

Next, preparation of a sample used for experiments, its sample loading, and its electrophoretic separations will be described. First, DNA size standards in every 100 bps each labeled with FITC (fluorescein isothiocyanate) (for example, such DNA size standards in every 100 bps each labeled with FITC may be purchased from "Bio-Rad Co.") were maintained at 4°C.

Thereafter, 2 μ m of a DNA ladder (size standard) solution was placed in the port 18b.

Sample loading into the agarose gel was carried out by applying 100 V to the capillary channel 16 through platinum electrodes mounted in the ports 18a and 18b.

In the following, experimental results obtained by applying electrophoretic separations to the sample introduced into the capillary channels 16 as described above will be described.

Fluorescence of FITC labeled DNA was detected by a system composed of an inverted fluorescence microscope (for example, "DIAPHOT-TMD; Nikon Co., Japan" may be used as such inverted fluorescence microscope) 100, an ICCD camera (for example, "C2400-8; Hamamatsu Photonics Co., Japan may be employed as such ICCD camera) 102 into which light from the inverted fluorescence microscope 100 is input, and a video camera 104 by which image signals output from the ICCD camera 102 are recorded.

The inverted fluorescence microscope 100 is composed of an xenon lamp 106, a band pass filter 108 allowing the light irradiated from the xenon lamp 106 to selectively pass through the same, a dichroic mirror 110 which allows the light transmitted from the band pass filter 108 to pass through the same and to output the light to the microchip 10 for capillary gel electrophoresis, and at the same time, reflects the light reflected by the microchip 10 for capillary gel electrophoresis, and a band pass filter 112 which allows the light reflected by the dichroic mirror 110 to selectively pass through the same and to input the light into the ICCD camera 102.

In this case, fluorescein was activated at 488 nm, emission was around 517 nm, and the image of electrophoresis was recorded by the video camera 104.

After the experiments, the image of electrophoresis was digitized by the use of an image analysis program (for example, "NIH image 1.62a" may be used as such image analysis program). Moreover, changes of electrophoresis with time was obtained by processing the digitized data with the use of a prescribed computer program.

FIG. 4 shows a scanning electron micrograph of the capillary channel 16 defined on the substrate 12 prepared from PDMS. As is clearly shown in FIG. 4, when the substrate 12 prepared from PDMS is used, a photoresist structure on the master which has been prepared from silicon can be transferred with high reproducibility.

The surface of a molded PDMS substrate 12 by using a master prepared from silicon is intrinsically hydrophobic which prevents capillary action between the capillary channel 16 and a gel solution.

Under the circumstances, in the present embodiment, a surface treatment with oxygen plasma was carried out for 2 minutes for the sake of filling the capillary channel 16 with an agarose solution as described above. As a result, a contact angle of water with respect to the PDMS substrate 12 changed from 108° to 32°, so that surface hydrophilicity of the substrate 12, i.e., surface hydrophilicity of the capillary channel 16 could be achieved.

Further, the cured PDMS substrate 12 could adhere to the PMMA surface plate 14 at ordinary temperatures without requiring any elaborate bonding manner. In addition, a master prepared from silicon containing the photoresist pattern can be utilized many times with just fluorocarbon treatment before molding.

Taking such fabricating processes into consideration, it is possible to fabricate the microchip 10 for capillary gel electrophoresis at extremely low cost, so that it is suitable for disposable use wherein the microchip is discarded after having been used for electrophoretic separations only one time.

Although agarose gel has been used as molecular sieves in the present embodiment, it has been generally known to be difficult to prepare homogeneously and stably a capillary

channel filled with a gel.

In this connection, gel instability during electrophoresis, i.e., bubble formation and clogging of pores in gels restricts electric field strength and separating performance of a gel.

In the present embodiment, however, the agarose can be easily introduced into the surface treated capillary channel 16 having hydrophilicity, and gelled, besides the whole operations can be completed within 10 minutes.

Moreover, neither bubble formation nor morphological change of agarose gel was observed during the electrophoresis wherein even 300 V was applied.

FIG. 5(A), (B), and (C) show situations of introduction and separation of DNA molecules, respectively.

More specifically, a sample plug could be formed by applying 100 V for 1 second (FIG. 5(A)).

Further, the separation process was visualized by the system shown in FIG. 3, and movements of bands could be clearly observed (FIG. 5(B) and FIG. 5(C)). The separation was attained by applying an electric field of 71.4 V/cm to 2.0% agarose gel wherein a TBE buffer solution is used. This electrical field strength is in a lower level than that of ordinary microfabricated capillary electrophoresis, i.e., several kV.

FIG. 6 indicates changes in electrophoresis of a microchip 10 for capillary gel with time wherein a capillary

channel 16 is filled with agarose gel containing DNA size standards of 100 bp to 1000 bp. As shown in the graph of FIG. 6, separation of DNA molecules ranging from 100 bp to 1000 bp was completed in 2 minutes which is ten to twenty times faster than conventional slab gel electrophoresis.

As explained, as to the microchip 10 for capillary gel electrophoresis prepared from PDMS according to the present invention in the above description, the microchip 10 for capillary gel electrophoresis can be easily fabricated in accordance with a simple and inexpensive molding method as well as a sealing method. It has been found that the microchip 10 for capillary gel electrophoresis is sufficiently available for DNA separation.

While in the above described preferred embodiment, the capillary channel 16 having a contour of a so-called I-shaped flow path has been defined on the substrate 12, the invention is not limited thereto as a matter of course. In this connection, for example, a capillary channel having a contour of a so-called cross-shaped flow path may be defined on the substrate 12. In the case where a capillary channel of a cross-shaped flow path has been defined on the substrate 12, four ports are bored on the surface plate 14 so as to correspond to four opposed ends of the capillary channel having a cross-shaped flow path, respectively.

Since the present invention has been constituted as described above, it is possible to provide a microchip for capillary gel electrophoresis which can be fabricated at low cost, and is suitable for disposable use, i.e., the microchip is discarded after having been used only one time in electrophoretic separation as well as to provide a method for fabricating such a microchip.

Furthermore, since the present invention has been constituted as described above, it is possible to provide a microchip for capillary gel electrophoresis by which it becomes possible to use a gel as molecular sieves, so that electrophoretic separations can be made at low voltage without employing any high voltage, and occurrence of diffusion and convection at the time of applying a voltage is prevented, whereby it is intended to simplify electrical equipment and a detection device, so that remarkable reduction of cost can be achieved as well as to provide a method for fabricating such a microchip.

Moreover, since the present invention has been constituted as described above, it is possible to provide a microchip by which a gel can be used as molecular sieves, whereby it is intended to improve a degree of separation, to reduce a separation distance, and to reduce a separation time, so that it becomes possible to improve separation performance in electrophoretic separations and to achieve an electrophoretic separation of high resolution as well as to provide a method for fabricating such a microchip.

It will be appreciated by those of ordinary skill in the art that the present invention can be embodied in other specific forms without departing from the spirit or essential characteristics thereof.

The presently disclosed embodiments are therefore considered in all respects to be illustrative and not restrictive. The scope of the invention is indicated by the appended claims rather than the foregoing description, and all changes that come within the meaning and range of equivalents thereof are intended to be embraced therein.

The entire disclosure of Japanese Patent Application No. 11-345050 filed on December 3, 1999 including specification, claims, drawings and summary are incorporated herein by reference in its entirety.

What is claimed is

1. A microchip for capillary gel electrophoresis, comprising:

a planar substrate prepared from a polymer material; and

a planer surface plate disposed on the top surface of said substrate;

a capillary channel constituting a flow path having a predetermined contour being defined on the top surface of said substrate; and

said capillary channel being sealed with said surface plate.

2. A microchip for capillary gel electrophoresis as claimed in claim 1, wherein

said substrate is prepared from PDMS (polydimethyl siloxane).

3. A microchip for capillary gel electrophoresis as claimed in claim 1, wherein

a surface of the capillary channel defined on the top surface of said substrate is made to be hydrophilic.

4. A microchip for capillary gel electrophoresis as claimed in claim 2, wherein

a surface of the capillary channel defined on the top surface of said substrate is made to be hydrophilic.

5. A microchip for capillary gel electrophoresis as claimed in claim 3, wherein

the hydrophilicity on the surface of the capillary channel defined on the top surface of said substrate is attained by oxidizing the surface of the capillary channel defined on the top surface of said substrate with oxygen plasma.

6. A microchip for capillary gel electrophoresis as claimed in claim 4, wherein

the hydrophilicity on the surface of the capillary channel defined on the top surface of said substrate is attained by oxidizing the surface of the capillary channel defined on the top surface of said substrate with oxygen plasma.

7. A method for fabricating a microchip for capillary gel electrophoresis, comprising:

a first treatment for printing a layout pattern of a capillary channel in a microchip for capillary gel electrophoresis on a transparent film to prepare a photolithographic mask;

a second treatment for forming a negative photoresist on a silicon wafer;

a third treatment for transferring the layout pattern printed on the mask prepared by said first treatment to the negative photoresist prepared by said second treatment, and developing the same to prepare a master;

a fourth treatment for treating the master prepared by said third treatment with fluorocarbon;

a fifth treatment for pouring a mixed solution consisting of a PDMS prepolymer and a curing agent over the master treated with fluorocarbon in said fourth treatment, and curing the same at a predetermined temperature for a predetermined period of time;

a sixth treatment for peeling a PDMS replica away from the master after completing the curing for a predetermined period of time in said fifth treatment, so that said PDMS replica is obtained as a substrate on which a capillary channel has been defined;

a seventh treatment for covering the substrate obtained in said sixth treatment by a surface plate, and attaching the latter to the former to seal said substrate with said surface plate; and

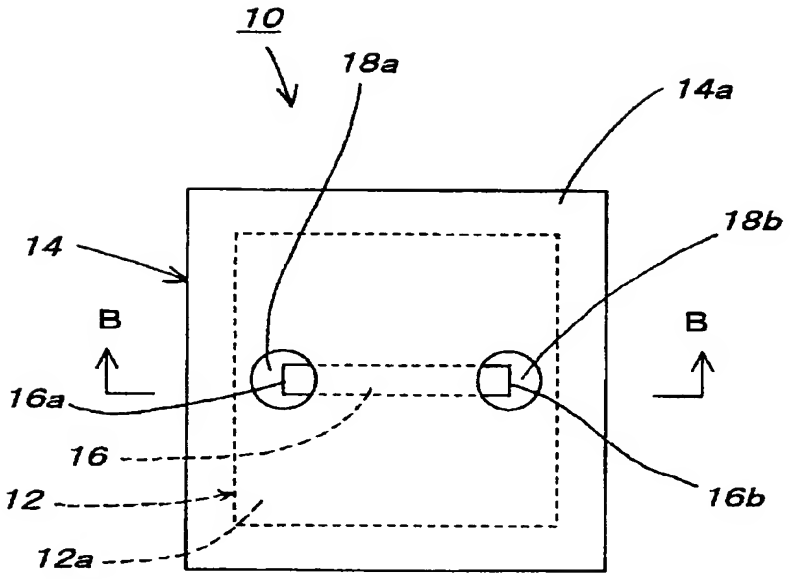
an eighth treatment for making a surface of the capillary channel defined on the PDMS substrate to which the surface plate has been attached hydrophilic.

8. A method for fabricating a microchip for capillary gel electrophoresis as claimed in claim 7, wherein

the treatment for making the surface of the capillary channel defined on the PDMS substrate in said eighth treatment hydrophilic is the one wherein the PDMS substrate to which the surface plate has been attached in said seventh treatment is oxidized with oxygen plasma, so that the capillary channel defined on said substrate is oxidized with oxygen plasma, whereby the surface of said capillary channel is made to be hydrophilic.

FIG. 1

(a)


10
18a
14a
14
18b
B
B
16a
16
12
12a
16b

(b)

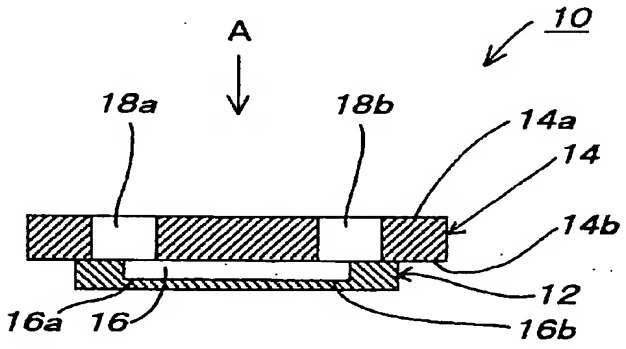

A
10
18a
18b
14a
14
14b
12
16a
16
16b

FIG. 2

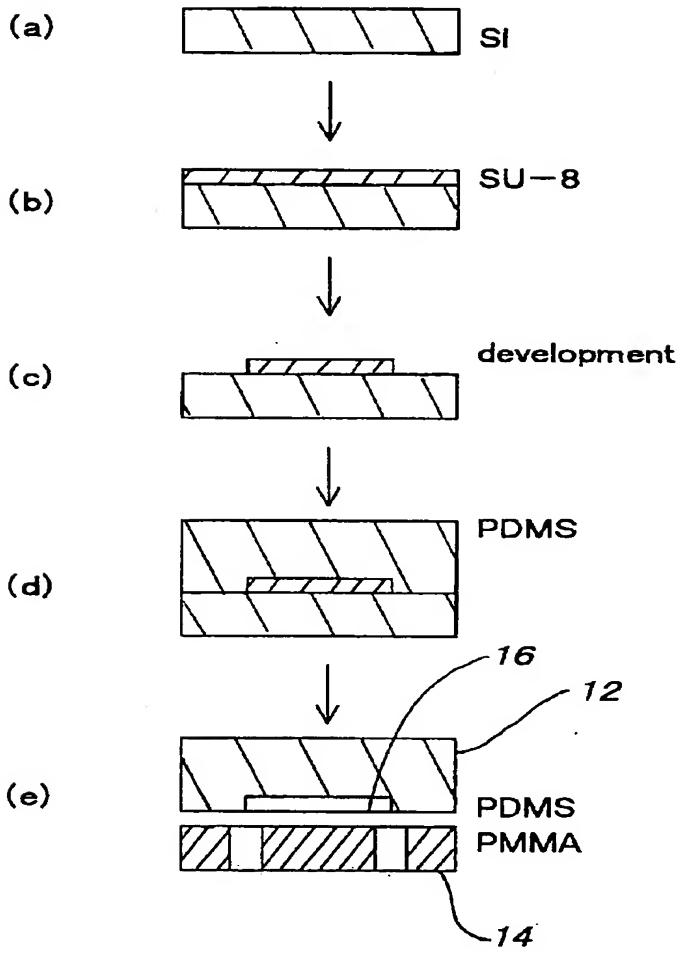


FIG. 3

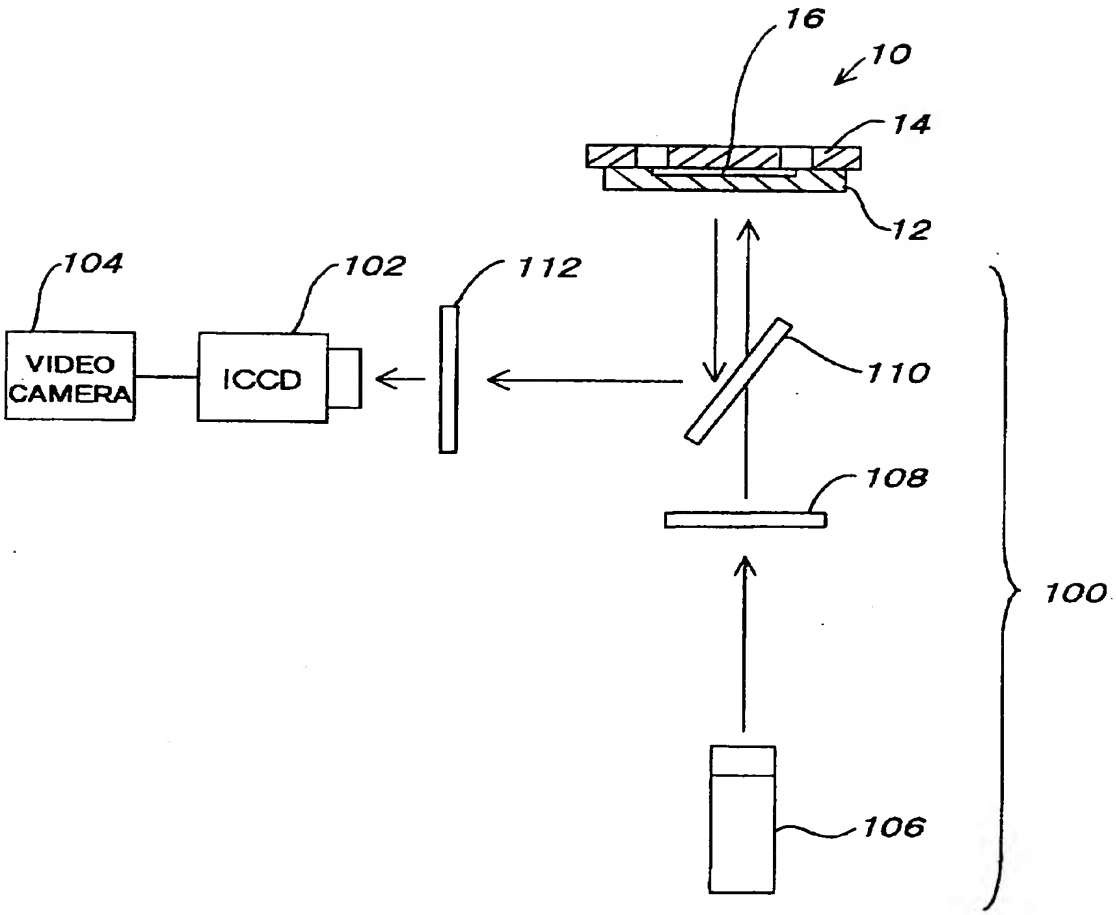


FIG. 4

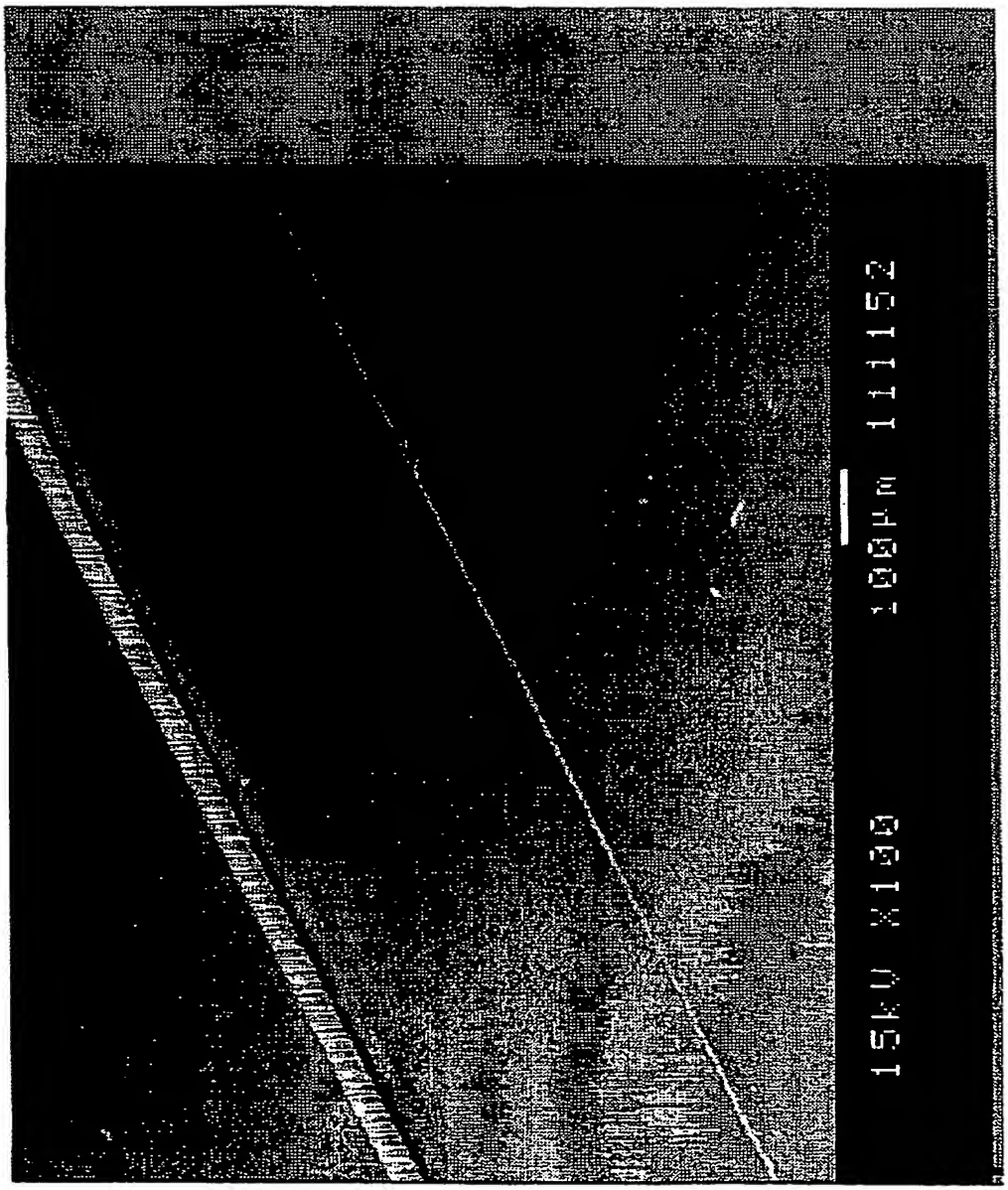
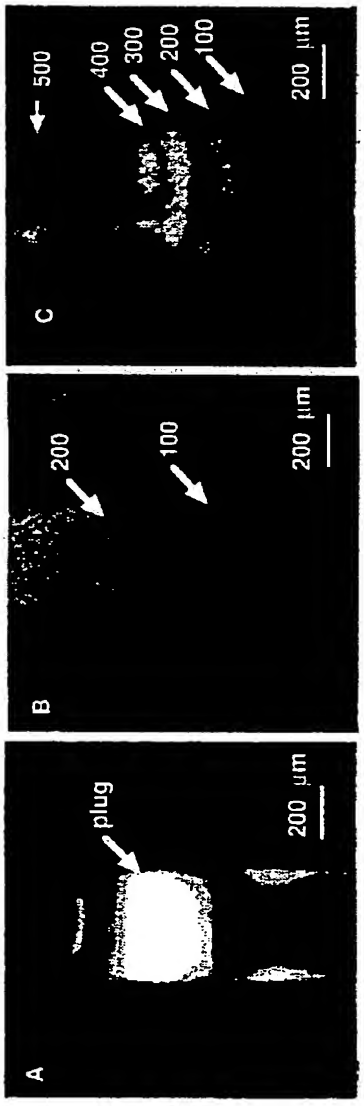


FIG. 5



(A) (B) (C)

FIG. 6

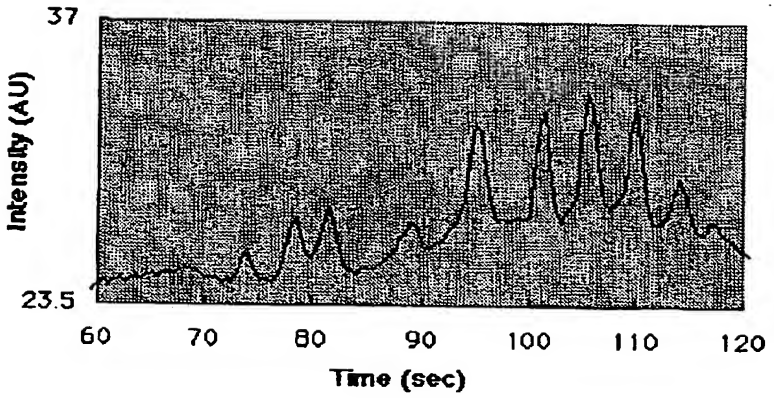


FIG. 7